

TOBACCO INDUSTRY RESEARCH COMMITTEE  
150 EAST FORTY SECOND STREET

NEW YORK 17, N.Y.

Renewal of Research Grant

#142 R 1  
Activated 1/1/57  
Cf #6 activated  
on 10/1/54 and  
renewed 10/1/55

Application ~~for Research Grant~~

Date: October 14, 1957

1. Name of Investigator: Paul D. Saltman, Ph.D.

2. Title: Assistant Professor  
Department of Biochemistry and Nutrition

3. Institution  
& Address: University of Southern California  
Los Angeles 7, California

4. Project or Subject:

Some aspects of amino acid metabolism in tobacco leaves. That several amino acids incorporate  $C^{14}O_2$  in the dark has been demonstrated in this laboratory using tobacco leaves. The biochemical pathways which are operative in the biosynthesis of these amino acids is at present unknown. It is proposed to investigate the enzymatic pathways mediating the metabolism of some of these important compounds.

5. Detailed Plan of Procedure (Use reverse side if additional space is needed):

During our first year of this grant we have been able to show conclusively that all of the serine produced during the dark fixation of  $CO_2$  by tobacco leaves is in the carboxyl carbon. Absolutely no radioactive carbon is to be found in the alpha or beta positions. This would seem to indicate a direct transformation from some three carbon precursor at the level of an organic acid. We have made extensive searches on our chromatograms to ascertain if any phosphorylated intermediate does indeed incorporate  $CO_2$  which would be on the pathway leading to serine. None was to be found except for phosphoglyceric acid. Radioactive pyruvate is also present and this could conceivably also lead into a three carbon precursor for serine.

Our attack now has shifted to the isolation of the enzymes involved in these reactions. It seems likely on the basis of the chromatographic data that the pools of these compounds present in the leaf are much too small to be measured. We therefore intend to work with homogenates with the hope that we can then isolate some of the intermediates in the pathway leading to serine biosynthesis.

We have also begun some work concerning the mechanisms of protein synthesis in tobacco and other plant as well as animal tissue. This work is now focused at the level of learning of the mechanisms involved in the activation of amino acids prior to their incorporation into the protein fraction of the leaf or cell. Mrs. Esther Allen is now actively engaged in this project and there seems to be a great promise for future developments along this line. Although this work was not in our original proposal, we felt it was of sufficient interest to warrant our investigations in this field.

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6. Budget Plan:

Salaries	\$5,000
Expendable Supplies	1,000
Permanent Equipment	1,000
Overhead	560
Other	
Total	\$7,560

7. Anticipated Duration of Work: 1 year

8. Facilities and Staff Available:

We have secured the cooperation of Dr. Fritz Went at California Institute of Technology who has placed the facilities of the Earhart Laboratories at our disposal for growing plants. We have a well equipped laboratory for biochemical investigations including: centrifuges, radioactive counting equipment, chromatographic equipment, cold rooms, spectrophotometer, etc.

Staff: Paul Saltman, Ph.D.

Esther Allen (pre-doctoral fellow)

Clyde Stitt (research assistant)

9. Additional Requirements:

10. Additional Information (Including relation of work to other projects and other sources of supply):

We have already published one paper concerning the work supported by the Tobacco Industries Research Committee concerning the metabolic pathways of the dark fixation carbon dioxide. We have also presented papers concerning further research along these lines at the last meeting of the Plant Physiological Society. The manuscripts for these papers are now in preparation and will be submitted to leading biochemical and plant physiological journals shortly.

Signature Paul Saltman  
Director of Project

A.V. Call  
Business Officer of the Institution  
President of the Board

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